

### **In the Claims**

This listing of claims will replace all prior versions, and listings, of claims in the application:

### **Listing of Claims**

Claims 1-32 (**canceled**).

Claim 33. (**new**) A method for preparing water-insoluble polysaccharides, comprising the steps of:

- (a) preparing a water solution of hydroxyl-containing polysaccharides;
- (b) adjusting a pH of the water solution of hydroxyl-containing polysaccharides to 2.5~7.5;
- (c) cross-linking the water solution of hydroxyl-containing polysaccharides by adding a poly-functional epoxy compound thereto, thereby forming a solution of cross-linked polysaccharides; and
- (d) shaping the solution of cross-linked polysaccharides into a water-insoluble polysaccharide.

Claim 34. (**new**)The method as claimed in claim 33, wherein the step (d) comprises pouring the solution of cross-linked polysaccharides into a mold, and yielding a film of water-insoluble polysaccharide after drying.

Claim 35. (**new**)The method as claimed in claim 33, wherein the step (d) comprises pouring the solution of cross-linked polysaccharides into a mold, and performing a freeze-drying to yield a porous water-insoluble polysaccharide.

Claim 36. (**new**)The method as claimed in claim 33, wherein the step (d) comprises adding organic solvent into the solution of cross-linked polysaccharides to precipitate cross-linked polysaccharides, and producing powders or sheets of water-insoluble polysaccharide by filtering.

Claim 37. **(new)**The method as claimed in claim 33, wherein the step (d) comprises squeezing the solution of cross-linked polysaccharides into a coagulant containing organic solvent to yield a fiber of water-insoluble polysaccharide.

Claim 38. **(new)**The method as claimed in claim 37, wherein the step (d) comprises intermittently squeezing the solution of cross-linked polysaccharides into a coagulant containing organic solvent to yield a sphere of water-insoluble polysaccharide.

Claim 39. **(new)**The method as claimed in claim 34, wherein the film of water-insoluble polysaccharide is applied in medicine or cosmetic after washing with water/organic solvent and distilled water and drying under vacuum.

Claim 40. **(new)**The method as claimed in claim 35, wherein the porous water-insoluble polysaccharide is applied in medicine or cosmetic after washing with water/organic solvent and distilled water and drying under vacuum.

Claim 41. **(new)**The method as claimed in claim 36, wherein the powders or sheets of water-insoluble polysaccharide is applied in medicine or cosmetic after washing with water/organic solvent and distilled water and drying under vacuum.

Claim 42. **(new)**The method as claimed in claim 37, wherein the fiber of water-insoluble polysaccharide is applied in medicine or cosmetic after washing with water/organic solvent and distilled water and drying under vacuum.

Claim 43. **(new)**The method as claimed in claim 38, wherein the sphere of water-insoluble polysaccharide is applied in medicine or cosmetic after washing with water/organic solvent and distilled water and drying under vacuum.

Claim 44. **(new)**The method as claimed in claim 33, wherein the hydroxyl-containing polysaccharide is selected from the group consisting of hyaluronic acid, carboxymethyl

cellulose, starch, alginate, chondroitin-4-sulfate, chondroitin-6-sulfate, xanthane gum, chitosan, pectin, agar, carrageenan, and guar gum.

Claim 45. **(new)**The method as claimed in claim 33, wherein, in the step (a), the hydroxyl-containing polysaccharides are dissolved in water to form a uniform water solution of hydroxyl-containing polysaccharides having a dry solid content of from 0.2 to 10% by weight.

Claim 46. **(new)**The method as claimed in claim 33, wherein the poly-functional epoxy compound is selected from the group consisting of 1,4-butanediol diglycidyl ether (BDDE), ethylene glycol diglycidyl ether (EGDGE), 1,6-hexanediol diglycidyl ether, polyethylene glycol diglycidyl ether, polypropylene glycol diglycidyl ether, polytetramethylene glycol diglycidyl ether, neopentyl glycol diglycidyl ether, polyglycerol polyglycidyl ether, diglycerol polyglycidyl ether, glycerol polyglycidyl ether, tri-methylolpropane polyglycidyl ether, pentaerythritol polyglycidyl ether, and sorbitol polyglycidyl ether.

Claim 47. **(new)**The method as claimed in claim 33, wherein, in the step (c), a molar equivalent ratio of the poly-functional epoxy compound to the hydroxyl-containing polysaccharide is in a range of between 0.1 and 8.0.

Claim 48. **(new)**The method as claimed in claim 47, wherein the molar equivalent ratio of the poly-functional epoxy compound to the hydroxyl-containing polysaccharide is 0.2 to 6.0.

Claim 49. **(new)**The method as claimed in claim 33, wherein the cross-linking is carried out at 10°C to 60°C for 10min to 12hrs.

Claim 50. **(new)**The method as claimed in claim 34, wherein the mold comprises ceramic, metal, or polymer, and the drying has a temperature between 25°C to 70°C.

Claim 51. **(new)**The method as claimed in claim 35, wherein the solution cross-linked polysaccharides is poured into the mold having a proper shape, and a porous water-insoluble polysaccharide having an interconnective pore structure is formed by the freeze-drying.

Claim 52. **(new)**The method as claimed in claim 36, wherein organic solvent is added into the solution of cross-linked polysaccharides to precipitate the cross-linked polysaccharides with stirring, and the powders or sheets of water-insoluble polysaccharide are produced by filtering.

Claim 53. **(new)**The method as claimed in claim 37, wherein the solution of cross-linked polysaccharides is intermittently squeezed into the coagulant containing organic solvent by a squeezer apparatus to produce the fiber of water-insoluble polysaccharide of a thickness of 50 $\mu$ m~1mm after drying.

Claim 54. **(new)**The method as claimed in claim 38, wherein the solution of cross-linked polysaccharides is intermittently squeezed into the coagulant containing organic solvent to produce the sphere of water-insoluble polysaccharide of a diameter of 50 $\mu$ m~1mm.

Claim 55. **(new)**The method as claimed in claim 53, wherein the coagulant containing organic solvent comprises water and organic solvent, and the organic solvent has a weight fraction between 60% and 100%.

Claim 56. **(new)**The method as claimed in claim 55, wherein the organic solvent is selected from the group consisting of 1, 4-dioxane, chloroform, methylene chloride, N, N-dimethylformamide (DMF), N, N-dimethylacetamide (DMAc), ethyl acetate, acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, and butanol.

Claim 57. **(new)**The method as claimed in claim 53, wherein the shaping is carried out at a coagulant temperature of 5°C to 60°C.

Claim 58. **(new)**The method as claimed in claim 39, wherein the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water, and the water/organic solvent has a temperature between 15°C and 50°C and the distilled water has a temperature between 25°C and 50°C, and the water/organic solvent comprises water and organic solvent, wherein the organic solvent is selected from the group consisting of acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination thereof, and the

organic solvent has a weight fraction between 50% and 100%, and acetone and alcohol are mixed with any ratio.

Claim 59. **(new)**The method as claimed in claim 40, wherein the porous water-insoluble polysaccharide is washed with water/organic solvent and distilled water, and the water/organic solvent has a temperature between 15°C and 50°C and the distilled water has a temperature between 25°C and 50°C, and the water/organic solvent comprises water and organic solvent, wherein the organic solvent is selected from the group consisting of acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination thereof, and the organic solvent has a weight fraction between 50% and 100%, and acetone and alcohol are mixed with any ratio.

Claim 60. **(new)**The method as claimed in claim 41, wherein the powders or sheets of water-insoluble polysaccharide are washed with water/organic solvent and distilled water, and the water/organic solvent has a temperature between 15°C and 50°C and the distilled water has a temperature between 25°C and 50°C, and the water/organic solvent comprises water and organic solvent, wherein the organic solvent is selected from the group consisting of acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination thereof, and the organic solvent has a weight fraction between 50% and 100%, and acetone and alcohol are mixed with any ratio.

Claim 61. **(new)**The method as claimed in claim 42, wherein the fiber of water-insoluble polysaccharide is washed with water/organic solvent and distilled water, and the water/organic solvent has a temperature between 15°C and 50°C and the distilled water has a temperature between 25°C and 50°C, and the water/organic solvent comprises water and organic solvent, wherein the organic solvent is selected from the group consisting of acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination thereof, and the organic solvent has a weight fraction between 50% and 100%, and acetone and alcohol are mixed with any ratio.

Claim 62. **(new)**The method as claimed in claim 43, wherein the sphere of water-insoluble polysaccharide is washed with water/organic solvent and distilled water, and the

water/organic solvent has a temperature between 15°C and 50°C and the distilled water has a temperature between 25°C and 50°C, and the water/organic solvent comprises water and organic solvent, wherein the organic solvent is selected from the group consisting of acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination thereof, and the organic solvent has a weight fraction between 50% and 100%, and acetone and alcohol are mixed with any ratio.

Claim 63. **(new)**The method as claimed in claim 58, wherein the film of water-insoluble polysaccharide is dried by hot air drying, radiation heating drying, or vacuum drying with a temperature below 60°C, after the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water.

Claim 64. **(new)**The method as claimed in claim 59, wherein the porous water-insoluble polysaccharide is dried by hot air drying, radiation heating drying, or vacuum drying with a temperature below 60°C, after the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water.

Claim 65. **(new)**The method as claimed in claim 60, wherein the powders or sheets of water-insoluble polysaccharide are dried by hot air drying, radiation heating drying, or vacuum drying with a temperature below 60°C, after the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water.

Claim 66. **(new)**The method as claimed in claim 61, wherein the fiber of water-insoluble polysaccharide is dried by hot air drying, radiation heating drying, or vacuum drying with a temperature below 60°C, after the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water.

Claim 67. **(new)**The method as claimed in claim 62, wherein the sphere of water-insoluble polysaccharide is dried by hot air drying, radiation heating drying, or vacuum drying with a temperature below 60°C, after the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water.

Claim 68. **(new)**A method for preparing water-insoluble polysaccharides, comprising the steps of:

- (a) preparing an organic solvent/water solution which contains an epoxy compound;
- (b) adjusting a pH of the organic solvent/water solution to 2.5~7.5;
- (c) adding shaped hydroxyl-containing polysaccharides into the pH-adjusted organic solvent/water solution; and
- (d) cross-linking the shaped hydroxyl-containing polysaccharides to form water-insoluble polysaccharides.

Claim 69. **(new)**The method as claimed in claim 68, wherein the epoxy compounds in the organic solvent/water solution have a weight fraction of about 0.05~2.0%.

Claim 70. **(new)**The method as claimed in claim 69, wherein the epoxy compounds in the organic solvent/water solution have a weight fraction of about 0.2~2.0%.

Claim 71. **(new)**The method as claimed in claim 68, wherein the epoxy compound is selected from the group consisting of 1,4-butanediol diglycidyl ether (BDDE), ethylene glycol diglycidyl ether (EGDGE), 1,6-hexanediol diglycidyl ether, polyethylene glycol diglycidyl ether, polypropylene glycol diglycidyl ether, polytetramethylene glycol diglycidyl ether, neopentyl glycol diglycidyl ether, polyglycerol polyglycidyl ether, diglycerol polyglycidyl ether, glycerol polyglycidyl ether, tri-methylolpropane polyglycidyl ether, pentaerythritol polyglycidyl ether, sorbitol polyglycidyl ether, and combination thereof.

Claim 72. **(new)**The method as claimed in claim 68, wherein the organic solvent in the organic solvent/water solution has a weight fraction of about 50~95%.

Claim 73. **(new)**The method as claimed in claim 72, wherein the organic solvent in the organic solvent/water solution has a weight fraction of about 70~90%.

Claim 74. **(new)**The method as claimed in claim 68, wherein organic solvent is a mixture of alcohol and acetone.

Claim 75. **(new)**The method as claimed in claim 68, wherein organic solvent is selected from the group consisting of 1, 4-dioxane, chloroform, methylene chloride, N, N-dimethylformamide (DMF), N, N-dimethylacetamide (DMAc), ethyl acetate, acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination thereof.

Claim 76. **(new)**The method as claimed in claim 68, wherein the hydroxyl-containing polysaccharide is selected from the group consisting of hyaluronic acid, carboxymethyl cellulose, starch, alginate, chondroitin-4-sulfate, chondroitin-6-sulfate, xanthane gum, chitosan, pectin, agar, carrageenan, guar gum, and combination thereof.

Claim 77. **(new)**The method as claimed in claim 68, wherein the shaped hydroxyl-containing polysaccharides comprise film, porosity, powder, sheet, fiber, or sphere.

Claim 78. **(new)**The method as claimed in claim 68, wherein the step (c) comprises pouring the solution of hydroxyl-containing polysaccharides into a mold to form a film of polysaccharide after drying, and adding the film of polysaccharide into the pH-adjusted organic solvent/water solution.

Claim 79. **(new)**The method as claimed in claim 68, wherein the step (c) comprises pouring the solution of hydroxyl-containing polysaccharides into a mold to form a porous polysaccharide by freeze-drying, and adding the porous polysaccharide into the pH-adjusted organic solvent/water solution.

Claim 80. **(new)**The method as claimed in claim 68, wherein the step (c) comprises adding organic solvent into the solution of hydroxyl-containing polysaccharides to precipitate hydroxyl-containing polysaccharides, after powders or sheets of polysaccharide are formed by filtering, adding the powders or sheets of polysaccharide into the pH-adjusted organic solvent/water solution.

Claim 81. **(new)**The method as claimed in claim 68, wherein the step (c) comprises intermittently squeezing the solution of hydroxyl-containing polysaccharides to form a sphere of



polysaccharide, and adding the sphere of polysaccharide into the pH-adjusted organic solvent/water solution.

Claim 82. **(new)**The method as claimed in claim 68, wherein the step (c) comprises squeezing the solution of hydroxyl-containing polysaccharides into a coagulant containing organic solvent to form a fiber of polysaccharide, and adding the fiber of polysaccharide into the pH-adjusted organic solvent/water solution.

Claim 83. **(new)**The method as claimed in claim 82, wherein the organic solvent is selected from the group consisting of 1, 4-dioxane, chloroform, methylene chloride, N, N-dimethylformamide (DMF), N, N-dimethylacetamide (DMAc), ethyl acetate, acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination thereof.

Claim 84. **(new)**The method as claimed in claim 68, wherein the cross-linking is carried out at 10°C to 60°C for 0.5hr to 12hrs.

Claim 85. **(new)**The method as claimed in claim 84, wherein the cross-linking is carried out at 25°C to 50°C.

Claim 86. **(new)**The method as claimed in claim 78, wherein the film of water-insoluble polysaccharide is applied in medicine or cosmetic after washing with water/organic solvent and distilled water and drying under vacuum.

Claim 87. **(new)**The method as claimed in claim 79, wherein the porous water-insoluble polysaccharide is applied in medicine or cosmetic after washing with water/organic solvent and distilled water and drying under vacuum.

Claim 88. **(new)**The method as claimed in claim 80, wherein the powders or sheets of water-insoluble polysaccharide are applied in medicine or cosmetic after washing with water/organic solvent and distilled water and drying under vacuum.

Claim 89. **(new)**The method as claimed in claim 81, wherein the fiber of water-insoluble polysaccharide is applied in medicine or cosmetic after washing with water/organic solvent and distilled water and drying under vacuum.

Claim 90. **(new)**The method as claimed in claim 82, wherein the sphere of water-insoluble polysaccharide is applied in medicine or cosmetic after washing with water/organic solvent and distilled water and drying under vacuum.

Claim 91. **(new)**The method as claimed in claim 86, wherein the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water, and the water/organic solvent has a temperature between 15°C and 50°C and the distilled water has a temperature between 25°C and 50°C, and the water/organic solvent comprises water and organic solvent, wherein the organic solvent is selected from the group consisting of acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination thereof, and the organic solvent has a weight fraction between 50% and 100%, and acetone and alcohol are mixed with any ratio.

Claim 92. **(new)**The method as claimed in claim 87, wherein the porous water-insoluble polysaccharide is washed with water/organic solvent and distilled water, and the water/organic solvent has a temperature between 15°C and 50°C and the distilled water has a temperature between 25°C and 50°C, and the water/organic solvent comprises water and organic solvent, wherein the organic solvent is selected from the group consisting of acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination thereof, and the organic solvent has a weight fraction between 50% and 100%, and acetone and alcohol are mixed with any ratio.

Claim 93. **(new)**The method as claimed in claim 88, wherein the powders or sheets of water-insoluble polysaccharide are washed with water/organic solvent and distilled water, and the water/organic solvent has a temperature between 15°C and 50°C and the distilled water has a temperature between 25°C and 50°C, and the water/organic solvent comprises water and organic solvent, wherein the organic solvent is selected from the group consisting of acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination

thereof, and the organic solvent has a weight fraction between 50% and 100%, and acetone and alcohol are mixed with any ratio.

Claim 94. **(new)**The method as claimed in claim 89, wherein the fiber of water-insoluble polysaccharide is washed with water/organic solvent and distilled water, and the water/organic solvent has a temperature between 15°C and 50°C and the distilled water has a temperature between 25°C and 50°C, and the water/organic solvent comprises water and organic solvent, wherein the organic solvent is selected from the group consisting of acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination thereof, and the organic solvent has a weight fraction between 50% and 100%, and acetone and alcohol are mixed with any ratio.

Claim 95. **(new)**The method as claimed in claim 90, wherein the sphere of water-insoluble polysaccharide is washed with water/organic solvent and distilled water, and the water/organic solvent has a temperature between 15°C and 50°C and the distilled water has a temperature between 25°C and 50°C, and the water/organic solvent comprises water and organic solvent, wherein the organic solvent is selected from the group consisting of acetone, methyl ethyl ketone (MEK), methanol, ethanol, propanol, isopropanol, butanol, and combination thereof, and the organic solvent has a weight fraction between 50% and 100%, and acetone and alcohol are mixed with any ratio.

Claim 96. **(new)**The method as claimed in claim 91, wherein the film of water-insoluble polysaccharide is dried by hot air drying, radiation heating drying, or vacuum drying with a temperature below 60°C, after the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water.

Claim 97. **(new)**The method as claimed in claim 92, wherein the porous water-insoluble polysaccharide is dried by hot air drying, radiation heating drying, or vacuum drying with a temperature below 60°C, after the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water.

Claim 98. **(new)**The method as claimed in claim 93, wherein the powders or sheets of water-insoluble polysaccharide are dried by hot air drying, radiation heating drying, or vacuum drying with a temperature below 60°C, after the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water.

Claim 99. **(new)**The method as claimed in claim 94, wherein the fiber of water-insoluble polysaccharide is dried by hot air drying, radiation heating drying, or vacuum drying with a temperature below 60°C, after the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water.

Claim 100. **(new)**The method as claimed in claim 95, wherein the sphere of water-insoluble polysaccharide is dried by hot air drying, radiation heating drying, or vacuum drying with a temperature below 60°C, after the film of water-insoluble polysaccharide is washed with water/organic solvent and distilled water.